Serican Journal of Medicine
2025, VOL. 2, NO. 1, 23193
https://doi.org/10.17161/sjm.v2i1.23193

Unveiling a non-canonical function of EZH2 in DNA damage repair and cancer progression

Andrea Cao[1] and Qingshu Meng[2]

[1]Brookfield East High School, Brookfield, WI 53005; [2]Department of Urology, Feinberg School of Medicine, Northwestern University, Chicago, IL 60611

Commentary

DNA damage repair (DDR) is an essential process for maintaining genomic stability [1, 2]. In cancer, however, this process is often dysregulated, contributing to uncontrolled cell proliferation and resistance to therapy [3]. Poly(adenosine diphosphate–ribose) polymerases (PARPs) are a family of pivotal enzymes in the DNA repair machinery, involving in several DNA repair pathways [4]. In which, PARP1 is the most abundant member and plays important roles in DDR and gene regulation [5]. When DNA damage occurs, PARP1 recognizes and binds to DNA lesions, then transfers ADP-ribose units from NAD+ to itself to form PAR chains. Repair complex including XRCC1 and Ligase III recognizes PAR chains as signals to recruit to damaged DNA and repair it [6]. Given its importance, PARPs have been evaluated as targets for cancer therapies, with inhibitors like olaparib and talazoparib showing promise in treating cancers harboring homologous recombination repair (HRR) mutations. Combination strategies aiming to enhance the therapeutic efficacy of PARP inhibitors, including dual targeting PARP1 and EZH2, are also under developing [7]. Enhancer of zeste homolog 2 (EZH2), on the other hand, is a histone methyltransferase traditionally known for its role in transcriptional suppression via methylation of histone H3 lysine 27 (H3K27). Recently, numerous studies revealed non-canonical functions of EZH2 in cancer development [8, 9]. It has been reported that EZH2 plays an important role in DDR by transcriptionally repressing some critical proteins in DDR pathways, such as RAD51 paralogs, SLFN11 and MAD2L2 [10-12]. Another research revealed that EZH2 can act as a transcriptional activator to activate genes involved in repair pathways [13]. More functions of EZH2 in DDR still are needed to be determined. A recent study published in *Science Advances* on November 27, 2024, titled "EZH2 directly methylates PARP1 and regulates its activity in cancer," uncovered a previous unidentified function of EZH2 in DNA damage repair and cancer progression by directly methylating PARP1 and fine-tuning its activity [14].

Meng et al. observed a direct interaction between EZH2 and PARP1 in Prostate cancer (PCa) cells and patient-derived xenograft tissue samples. Interestingly, EZH2 depletion and catalytic inhibition remarkably enhance PARP1 activity without altering its transcript or protein expression levels, which indicated a non-canonical methyltransferase-dependent manner of EZH2 in repressing PARP1 activity. Through posttranslational modification analysis by mass spectrometry and site-directed mutagenesis assay, Meng et al. demonstrated that EZH2 directly methylates PARP1 at lysine 607. The study revealed that PARP1 is a novel non-histone substrate of EZH2, and PARP1 lysine methylation significantly alters PARP1's enzymatic activity.

Next, Meng et al. performed bioluminescent NAD detection assay, *in vivo* and *in vitro* PARP1 autoPARylation, proximity ligation assay (PLA) and Comet assay to reveal how EZH2-mediated PARP1 methylation affects its role in DDR. Results indicated that EZH2-mediated PARP1 methylation protects cells against NAD+ over-consumption, represses PARP1 autoPARylation, reduces PARylation-dependent XRCC1 recruitment and finally downregulates DNA damage repair. This finding expands the functional repertoire of EZH2 in DDR.

In addition to DNA damage repair, PARP1 is also involved in transcriptional regulation [15]. The study further explores how EZH2-mediated methylation

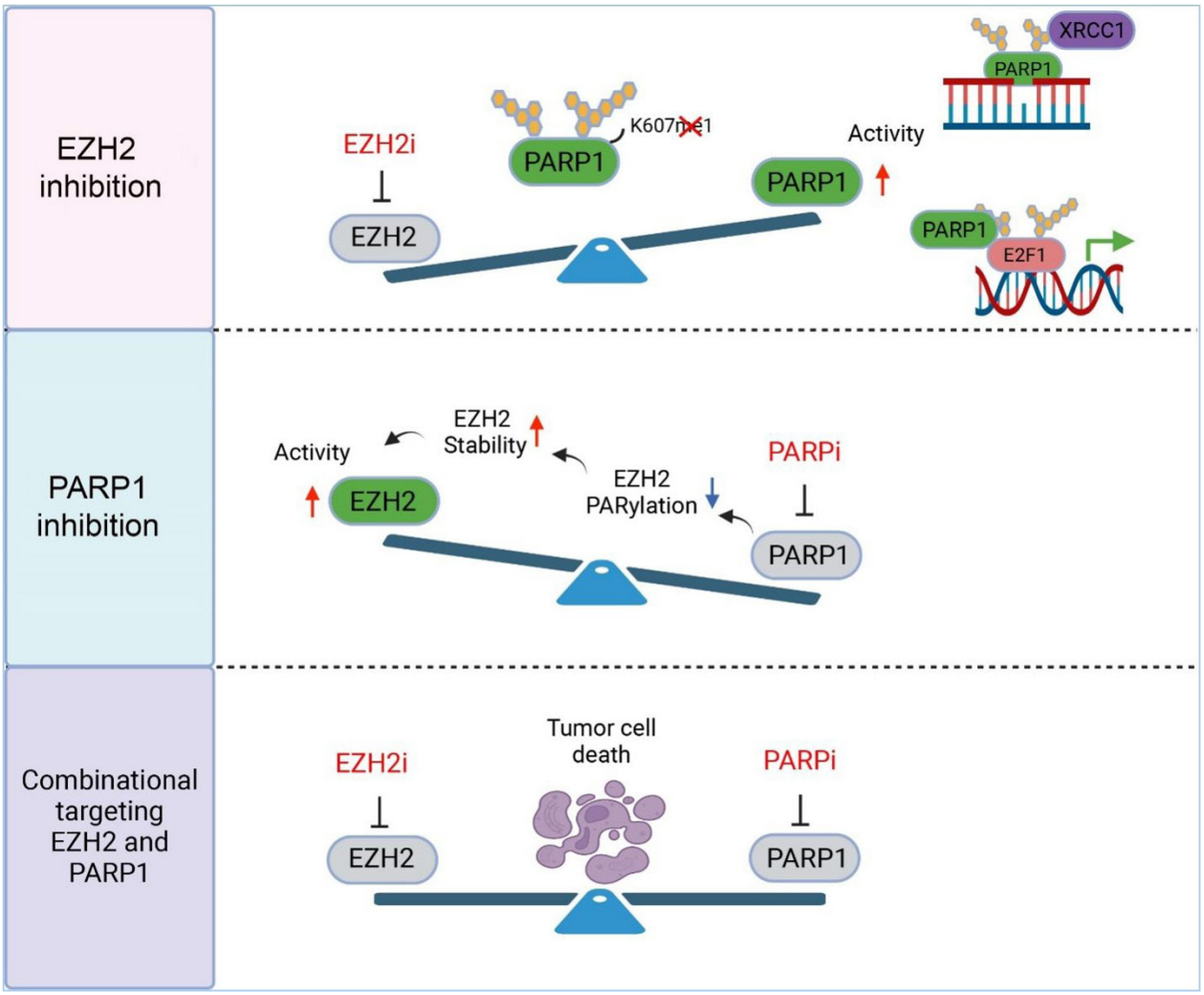
ARTICLE HISTORY
Received: Dec. 10, 2024
Revised: Jan. 2, 2025
Accepted: Jan. 14, 2025

KEYWORDS
EZH2, DNA damage repair, cancer progression, PARP inhibitor, H3K27

EDITED BY
Jindan Yu

REVIEWED BY
Xiaodong Lu, Ling Cai

Figure 1. Schematic diagram for bidirectional regulation of EZH2-PARP1 axis and combination strategy of EZH2 and PARP1 inhibition. When only targeting EZH2, EZH2i inhibits PARP1 lysine methylation and increases its catalytic activity, upregulates the recruitment of XRCC1 to DNA lesions and associated DNA damage repair, enhances PARP1-E2F1 interaction and E2F1 transcription factor activity, hence promoting PARP1-mediated tumor progression. While targeting PARP1 alone, PARPi inhibits EZH2 PARylation and subsequent degradation, thereby promoting EZH2-mediated gene silencing and cancer progression. Only combinational targeting of EZH2 and PARP1 can keep the "seesaw" balanced and sufficiently induce tumor cell death.



affects PARP1's transcriptional functions. By impairing the interaction between PARP1 and E2F1, a renowned transcription factor, the methylation inhibits E2F1's transcriptional activity. This finding links EZH2 activity to the regulation of PARP1-mediated oncogenic pathways, providing insight into how these proteins collaborate to promote cancer progression.

The study demonstrated that EZH2 directly methylates PARP1 and downregulates its activity, and also validated the previous reported PARP1-mediated EZH2 PARylation and activity alteration in PCa cells [16]. The authors use a "seesaw" model to describe this bidirectional regulation of EZH2-PARP1 axis (Figure 1). As the last part, the authors highlight the synergistic effects of combining EZH2 and PARP1 inhibitors in suppressing prostate cancer growth. This observation underscores the therapeutic potential of dual targeting in cancers with EZH2 overexpression and PARP1 dependence, offering a compelling rationale for combinatorial approaches in clinical settings.

One of the strengths of this work lies in its ability to elucidate a novel regulatory mechanism in cancer biology. The direct methylation of PARP1 by EZH2 represents an important addition to the growing list

of non-histone substrates of EZH2, broadening our understanding of its roles in tumor progression. Additionally, the study also excels in linking mechanistic insights to therapeutic strategies. By demonstrating the efficacy of combining EZH2 and PARP1 inhibitors, the authors translate their findings into a preclinical setting, bridging the gap between basic and translational research.

Despite its strengths, the study leaves some questions unanswered. For instance, the molecular basis of how EZH2 recognizes PARP1 as a substrate remains unclear. Structural studies elucidating the interaction between these two proteins could provide deeper insights into the specificity of this modification.

In summary, the findings presented in this study mark a new contribution to our understanding of the molecular underpinnings of cancer. By identifying EZH2 as a regulator of PARP1 activity, the authors not only uncover a novel mechanism of DNA repair dysregulation but also provide a strong rationale for combinatorial therapies targeting these two proteins. As research continues to unravel the intricate networks governing cancer progression, studies like this pave the way for innovative and effective treatments that could be exploited clinically.

Reference

1. Lombard DB, Chua KF, Mostoslavsky R, Franco S, Gostissa M, Alt FW: **DNA repair, genome stability, and aging**. *Cell* 2005, **120**(4):497-512. doi: 10.1016/j.cell.2005.01.028.
2. Tubbs A, Nussenzweig A: **Endogenous DNA Damage as a Source of Genomic Instability in Cancer**. *Cell* 2017, **168**(4):644-656. doi: 10.1016/j.cell.2017.01.002. PMC6591730.
3. Groelly FJ, Fawkes M, Dagg RA, Blackford AN, Tarsounas M: **Targeting DNA damage response pathways in cancer**. *Nat Rev Cancer* 2023, **23**(2):78-94. doi: 10.1038/s41568-022-00535-5.
4. Gibson BA, Kraus WL: **New insights into the molecular and cellular functions of poly(ADP-ribose) and PARPs**. *Nat Rev Mol Cell Biol* 2012, **13**(7):411-424. doi: 10.1038/nrm3376.
5. Huang D, Kraus WL: **The expanding universe of PARP1-mediated molecular and therapeutic mechanisms**. *Mol Cell* 2022, **82**(12):2315-2334. doi: 10.1016/j.molcel.2022.02.021. PMC9232969.
6. Ray Chaudhuri A, Nussenzweig A: **The multifaceted roles of PARP1 in DNA repair and chromatin remodelling**. *Nat Rev Mol Cell Biol* 2017, **18**(10):610-621. doi: 10.1038/nrm.2017.53. PMC6591728.
7. Li X, Wang C, Li S, Yin F, Luo H, Zhang Y, Luo Z, Chen Y, Wan S, Kong L *et al*: **Dual target PARP1/EZH2 inhibitors inducing excessive autophagy and producing synthetic lethality for triple-negative breast cancer therapy**. *Eur J Med Chem* 2024, **265**:116054. doi: 10.1016/j.ejmech.2023.116054.
8. Park SH, Fong KW, Mong E, Martin MC, Schiltz GE, Yu J: **Going beyond Polycomb: EZH2 functions in prostate cancer**. *Oncogene* 2021, **40**(39):5788-5798. doi: 10.1038/s41388-021-01982-4. PMC8487936.
9. Zimmerman SM, Lin PN, Souroullas GP: **Non-canonical functions of EZH2 in cancer**. *Front Oncol* 2023, **13**:1233953. doi: 10.3389/fonc.2023.1233953. PMC10473085.
10. Zeidler M, Varambally S, Cao Q, Chinnaiyan AM, Ferguson DO, Merajver SD, Kleer CG: **The Polycomb group protein EZH2 impairs DNA repair in breast epithelial cells**. *Neoplasia* 2005, **7**(11):1011-1019. doi: 10.1593/neo.05472. PMC1502020.
11. Gardner EE, Lok BH, Schneeberger VE, Desmeules P, Miles LA, Arnold PK, Ni A, Khodos I, de Stanchina E, Nguyen T *et al*: **Chemosensitive Relapse in Small Cell Lung Cancer Proceeds through an EZH2-SLFN11 Axis**. *Cancer Cell* 2017, **31**(2):286-299. doi: 10.1016/j.ccell.2017.01.006. PMC5313262.
12. Karakashev S, Fukumoto T, Zhao B, Lin J, Wu S, Fatkhutdinov N, Park PH, Semenova G, Jean S, Cadungog MG *et al*: **EZH2 Inhibition Sensitizes CARM1-High, Homologous Recombination Proficient Ovarian Cancers to PARP Inhibition**. *Cancer Cell* 2020, **37**(2):157-167 e156. doi: 10.1016/j.ccell.2019.12.015. PMC7155421.
13. Liao Y, Chen CH, Xiao T, de la Pena Avalos B, Dray EV, Cai C, Gao S, Shah N, Zhang Z, Feit A *et al*: **Inhibition of EZH2 transactivation function sensitizes solid tumors to genotoxic stress**. *Proc Natl Acad Sci U S A* 2022, **119**(3). doi: 10.1073/pnas.2105898119. PMC8784159.
14. Meng Q, Shen J, Ren Y, Liu Q, Wang R, Li Q, Jiang W, Wang Q, Zhang Y, Trinidad JC *et al*: **EZH2 directly methylates PARP1 and regulates its activity in cancer**. *Sci Adv* 2024, **10**(48):eadl2804. doi: 10.1126/sciadv.adl2804. PMC11601213.
15. Schiewer MJ, Knudsen KE: **Transcriptional roles of PARP1 in cancer**. *Mol Cancer Res* 2014,

12(8):1069-1080. doi: 10.1158/1541-7786.MCR-13-0672. PMC4134958.

16. Yamaguchi H, Du Y, Nakai K, Ding M, Chang SS, Hsu JL, Yao J, Wei Y, Nie L, Jiao S *et al*: **EZH2 contributes to the response to PARP inhibitors through its PARP-mediated poly-ADP ribosylation in breast cancer**. *Oncogene* 2018, **37**(2):208-217. doi: 10.1038/onc.2017.311. PMC5786281.